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TITLE: Genes Associated with Food Allergy and Eosinophilic Esophagitis

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<b>14. ABSTRACT</b>  The ingestion of food antigens plays an essential role in the development of eosinophilic esophagitis (EE) as total removal of dietary antigens by using an amino acid based oral formula improves clinical symptoms and esophageal histology in 98% of patients with EE within a month. EE is thought to be mediated by both IgE and non-IgE mediated food allergy. In this study we are particularly interested in identifying genes in EE linked to a significant complication of EE namely esophageal stricture formation. This study focuses on increasing our understanding of two genes (TGF- $\beta$ and acidic chitinase) associated with remodeling and stricture formation in the esophagus in EE. The importance of TGF- $\beta$ and acidic chitinase to the development of egg induced remodeling of the esophagus is being studied in a mouse model in which either TGF- $\beta$ signaling or acidic chitinase activity is neutralized. In the 6 months since the mouse protocol was approved we have completed breeding of Smad3 deficient mice and investigated whether TGF- $\beta$ is important in egg induced remodeling in a mouse model of EE in Smad-3 deficient mice (deficient in TGF- $\beta$ signaling) compared to WT mice. Results will be available in 6 months.						
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## **Introduction:**

The ingestion of food antigens play s an essential role in the development of eosinophilic esophagitis (EE) as total removal of dietary antigens by using an amino acid based oral formula improves clinical symptoms and esophageal histology in 98% of patients with EE within a month. EE is thought to be mediated by both IgE and non-IgE mediated food allergy. In this study we are particularly interested in identifying genes in EE linked to a significant complication of EE namely esophageal stricture formation. This study focuses on increasing our understanding of two genes TGF- $\beta$  (transforming growth factor- $\beta$ ) and acidic chitinase associated with remodeling and stricture formation in the esophagus in EE. The importance of TGF- $\beta$  and acidic chitinase to the development of egg induced remodeling of the esophagus is being studied in a mouse model in which either TGF- $\beta$  signaling is inhibited or acidic chitinase activity is neutralized. TGF- $\beta$  will be inactivated in studies of Smad3 deficient mice (essential for TGF- $\beta$  signaling) and chitinase will be inactivated in studies of mice administered an anti-chitinase Ab.

## **Body:**

This proposal outlines 4 tasks to be completed during the three year proposal. In year 1 of this proposal we have worked on task 1 and task 2 as planned in our original proposal.

### **1. Task 1:** Breeding of Smad3 deficient mice (month 1-3)

The first year of this award was from July 1, 2010 to June 30, 2011.

Month 1-6 was spent obtaining revisions to the animal subject's protocol requested by DOD. We received an approval letter from DOD permitting us to start working on the project on January 3, 2011 in an e-mail from Ms Kathleen Dennis (Administrative Assistant ACURO; Kathleen.Dennis1@us.army.mil) which had an attached approval letter from Alec Hail DVM, Director ACURO.

Thus, starting our task 1 was delayed by 6 months from month 1-3, to months 6-9.

We have nevertheless completed task 1 breeding of Smad3 deficient mice and started to use them in experiments proposed for task 2.

### **2. Task 2:** Mouse model of egg induced EE (WT vs Smad3 deficient) (month 4-16)

Task 2 (a) Exposure of WT and Smad3 deficient mice to OVA allergen

The experiments with the WT and Smad3 deficient mice have been performed in 16/48 mice proposed to be studied in this task thus far. The experiments in the 16 mice (four groups of 4 mice ; WT no OVA; WT OVA; Smad3ko no OVA; Smad3ko OVA) finished on July 13, 2011 and esophageal specimens have been processed for immunohistology (task 2b-g).

The remaining 32 mice will be studied in two groups of 16 mice. Each of these two 16 mouse experiments will end before month 16 as originally proposed.

### Task 2 (b-g)

Quantitating fibrosis (2b), basal zone hyperplasia (2c), blood vessels (2d), eosinophils (2e), TGF- $\beta$ + cells (2f), and pSmad+ cells (2g) has just started on the first 16 mice and this analysis will be completed by month 16 as originally proposed.

Because of the 6 month delay in obtaining animal subjects approval, the completion of Task 2 (b-g) in the remaining 32/48 mice will be delayed by 6 months to month 22. Although this will delay completion of task 2 (b-g), this will not delay task 3 (perform during month 16-28) or task 4 (month 33-36).

## **Key Research Accomplishments:**

- Breeding of Smad3 deficient mice
- Experiment with OVA induced EE in WT vs Smad3 deficient mice (33% of mice completed)
- Esophagus from 12 mice processed for immunohistology

## **Reportable Outcomes:**

There are currently no reportable outcomes in the first 6 months of working on this project.

**Conclusion:**

There are currently no conclusions.

**References:**

None.

**Appendices:**

None.

**Manuscripts/Reprints, Abstracts:**

None.